

INVESTIGATION OF SOME OF THE THERMODYNAMICAL QUANTITIES OF PROTEINS BY STATISTICAL MECHANICAL METHODS

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Abstract

In this study, variations with respect to temperature of the increments of enthalpy ΔH and Gibbs energy ΔG , arising in the dissolution of proteins in water, have been investigated by the methods of statistical thermodynamics. In this formalism, effective electric field E and total dipole moment M are taken as thermodynamical variable. In obtaining the free energy the partition function given by A. Bakk, J.S. Høye and A. Hansen; Physica A, 304, (2002), 355-361 has been used in a modified form. In the constructed semi-phenomenological theory, the experimental data are taken from the study of Privalov [1, 2] and the relevant parameters have been found by fitting to the experimental curves. The variations of the increments of enthalpy ΔH and Gibbs energy ΔG have been investigated in the temperature range 265 – 350K.

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1 Introduction

Proteins is the common name of the complex macro molecules which are formed by gathering of a great number amino-acids and which play unremitting roles in the life times of all of the living creatures. There are approximately twenty different amino-acids which take place in the structure of the proteins and which are chain molecules. The order or arrangement of this twenty units is sensitive and definite. This order, determines the nature of the protein and as a result identifies its function [3]. Each protein molecule is formed by on original combination and consecutive placing of different numbers of amino-acids.

Proteins under their own natural physiological conditions, spontaneously open out from one dimensional structures to three dimensional structures in the time intervals starting from milliseconds and getting to minutes [4, 5]. The three dimensional structures of the proteins depend on various environmental factors in the cell. Unfolded proteins return to their natural states when the environmental factors are removed exhibiting the feature that the three dimensional structure of a protein solely depends on its amino-acid ordering information. This situation gives the hope to some research workers that protein folding could be reconstructed by computer simulation [6]. But the difficulty of this simulation even for small proteins is in view.

In this study, the model belonging to the proteins is presented in section 2. In section 3, the thermodynamical quantities concerning the variation of the increments of additional enthalpy ΔH and Gibbs energy ΔG with respect to temperature have been expressed and these parameters are related to the parameters of the proteins with the help of statistical thermodynamics. At the end, from the curves belonging to the increments of enthalpy and Gibbs energy, the conclusions relevant to the structures of the proteins are presented.

2 Model

A simple model of protein unfolding has been developed by Bakk [7]. Bakk has started the model by choosing the water molecules as classical electric dipoles. he has demanded that non polar dissolution should include important physical properties in order to model the effect on the folding of the proteins. In the model used, in the medium of the formed electric field, the

folding and unfolding of the proteins have been taken as the basis. Here, the electric field is not an external field but has been used in the modelling of the ice-like behavior exhibited by the water molecules around the non polar surfaces. The electric field is a result of the effective behavior of the non polar dissolvent applied to the protein unfolding.

The modified partition function of the protein system could be taken as [7]:

$$Z(T, E, \mu) = \exp[\exp(\frac{\mu}{kT})z(T, E, N = 1)]. \quad (1)$$

Here, μ represents the chemical potential and the term $\zeta = \exp(\beta\mu)$ corresponding to the pH of the system has been introduced to the partition function. $z(T, E, N = 1) = 4\pi \exp^{-\frac{\beta b m^2}{2}} \frac{1}{\beta \epsilon_e} \sinh \beta \epsilon_e$ indicates single particle partition function. Where effective energy is given by $\epsilon_e = \epsilon + b < m > [7]$. On the other hand average dipole moment has the form $< m > = L(\beta \epsilon_e)$.

3 Variations of the Enthalpy and Gibbs Energy Increments with Respect to Temperature

Taking electric field E and total dipole moment M as thermodynamical variables, the first law of thermodynamics could be written for the proteins in the form:

$$dU = TdS - MdE. \quad (2)$$

In this case, change in the enthalpy is;

$$dH = TdS + EdM. \quad (3)$$

Enthalpy and entropy increments could also be written by taking the total differentials of enthalpy $H = H(S, M)$ and entropy $S = S(T, M)$ which leads to:

$$dH = (\frac{\partial H}{\partial S})_M dS + (\frac{\partial H}{\partial M})_S dM \quad (4)$$

$$dS = (\frac{\partial S}{\partial T})_M dT + (\frac{\partial S}{\partial M})_T dM. \quad (5)$$

After substituting Eq.(4) and Eq.(5) in Eq.(3)

$$dH = (\frac{\partial H}{\partial S})_M (\frac{\partial S}{\partial T})_M dT + [(\frac{\partial H}{\partial S})_M (\frac{\partial S}{\partial M})_T + (\frac{\partial H}{\partial M})_S] dM \quad (6)$$

has been obtained.

Writing down the expressions $T = (\frac{\partial H}{\partial S})_M$, $E = (\frac{\partial H}{\partial M})_S$ and $(\frac{\partial S}{\partial T})_M = \frac{\Delta C_M}{T}$ in Eq.(6) and then integrating one could get for the enthalpy increment:

$$\Delta H = \int_{T_i}^T C_M dT + \int_{M(T_i)}^{M(T)} [E + T(\frac{\partial S}{\partial M})_T] dM. \quad (7)$$

Using the first law of thermodynamics for the proteins which has been given by Eq.(2), change in the Gibbs energy

$$dG = -SdT + EdM \quad (8)$$

could be written down. With a similar approach, taking the total differentials of the Gibbs energy $G = G(T, M)$ and total dipole moment $M = M(T, E)$ and then substituting then in Eq.(8) together with the expressions $E = (\frac{\partial G}{\partial M})_T$ and $-S = (\frac{\partial G}{\partial T})_M$ leads after integration to the expression which gives the Gibbs energy increment:

$$\Delta G = - \int_{T_i}^T SdT + \int_{M(T_i)}^{M(T)} EdM. \quad (9)$$

The free energy of the system has been obtained from the relation $F = -kT \ln Z$ giving:

$$F = - \frac{4 \exp(-\beta(\frac{1}{2}bm^2 - \mu))\pi \sinh(\beta\epsilon_e)}{\beta^2\epsilon_e}. \quad (10)$$

The thermodynamical quantities of the system has been determined using the entropy relation $S = -(\frac{\partial F}{\partial T})_E$ which in turn leads to:

$$S = \frac{2 \exp(-\beta(\frac{1}{2}bm^2 - \mu))k\pi}{\beta\epsilon_e} \times \{(-2\epsilon_e\beta \cosh(\beta\epsilon_e) + (4 + \beta bm^2 - 2\beta\mu) \sinh(\beta\epsilon_e))\}. \quad (11)$$

The total dipole moment has been calculated from the expression $M = (\frac{\partial F}{\partial E})_T$ resulting in the following:

$$M = \frac{4 \exp(-\beta(\frac{1}{2}bm^2 - \mu))\pi}{\beta^2\epsilon_e^2} \times \{\epsilon_e\beta \cosh(\beta\epsilon_e) - \sinh(\beta\epsilon_e)\}. \quad (12)$$

The energy of the system is given by $U = -\frac{1}{Z} \frac{\partial Z}{\partial \beta}$ and when the partition function is substituted one obtains:

$$U = \frac{\exp(-\beta(\frac{1}{2}bm^2 - \mu))}{\beta^2\epsilon_e} \times \{(-4\pi\epsilon_e\beta \cosh(\beta\epsilon_e) + 2\pi(2 + \beta bm^2 - 2\beta\mu) \sinh(\beta\epsilon_e))\}. \quad (13)$$

Additional heat capacity at effective field has been calculated by making use of the equation $\Delta C_E = -k\beta^2(\frac{\partial U}{\partial \beta})_E$ which gives:

$$\Delta C_E = \frac{\exp(-\beta(\frac{1}{2}bm^2 - \mu))k\pi}{\beta\epsilon_e} \times \{-8\epsilon_e\beta \cosh(\beta\epsilon_e) - (4\epsilon_e\beta(\beta bm^2 + 2\beta\mu)) \cosh(\beta\epsilon_e) + (8 + \beta(b^2m^4\beta - 8\mu)) \sinh(\beta\epsilon_e) - \beta 4bm^2(\beta\mu - 1) \sinh(\beta\epsilon_e) + 4\beta^2(\epsilon_e^2 + \mu^2) \sinh(\beta\epsilon_e)\}. \quad (14)$$

Using the expression $\Delta C_M - \Delta C_E = -T(\frac{\partial M}{\partial E})_T[(\frac{\partial E}{\partial T})_M]^2$ which has been obtained for additional heat capacity at constant total dipole moment;

$$\Delta C_M = \frac{\exp(-\beta(\frac{1}{2}bm^2 - \mu))k\pi}{\beta\epsilon_e} \times \{-4\epsilon_e\beta(2 + \beta bm^2 - 2\beta\mu) \cosh(\beta\epsilon_e) + [8 + \beta(b^2m^4\beta - 8\mu - 4bm^2(\beta\mu - 1) + 4\beta(\epsilon_e^2 + \mu^2))] \sinh(\beta\epsilon_e) - \frac{k^2\beta^2}{(-2\beta\epsilon_e \cosh(\beta\epsilon_e) + (2 + \beta^2\epsilon_e^2) \sinh(\beta\epsilon_e))} \times [\epsilon_e\beta(4\beta bm^2 - 2\beta\mu) \cosh(\beta\epsilon_e) - (4 + \beta(bm^2 + 2\beta\epsilon_e - 2\mu)) \sinh(\beta\epsilon_e)]^2\} \quad (15)$$

has been calculated.

Another quantity $(\frac{\partial S}{\partial M})_T$ when calculated gives:

$$(\frac{\partial S}{\partial M})_T = -k\beta\epsilon_e. \quad (16)$$

When Eq.(15) and Eq.(16) are substituted in Eq.(7), after integration gives for the enthalpy increment:

$$\begin{aligned}
\Delta H = & \frac{2\pi \exp(-\frac{bm^2(T_t+2T)}{2kT_tT})}{\epsilon_e^2} \times \{T_t \exp(\frac{bm^2(T_t+2T)+2T\mu}{2kT_tT}) \times \\
& [2\epsilon_e(bm+\epsilon-k\epsilon+(k-1)\epsilon_e) \cosh(\frac{\epsilon_e}{kT_t}) + k(2(k-1)T_t(\epsilon-\epsilon_e) - \\
& bm(2T_t+m\epsilon_e)+2\epsilon_e\mu) \sinh(\frac{\epsilon_e}{kT_t})] + T \exp(\frac{bm^2T+T_t\mu}{2kT_tT}) \times \\
& [-2\epsilon_e(bm+\epsilon-k\epsilon+(k-1)\epsilon_e) \cosh(\frac{\epsilon_e}{kT}) + k(-2(k-1)T(\epsilon-\epsilon_e) + \\
& bm(2T+m\epsilon_e)-2\epsilon_e\mu) \sinh(\frac{\epsilon_e}{kT})] \}. \quad (17)
\end{aligned}$$

Similarly, substituting Eq.(11) into Eq.(9), after integration and writing down Eq.(12) in this expression, one obtains for the Gibbs energy increment:

$$\begin{aligned}
\Delta G = & \frac{4k\pi \exp(-\frac{bm^2(T_t+T)}{2kT_tT})}{\epsilon_e^2} \times \{T_t \exp(\frac{\frac{bm^2}{T} + \frac{2\mu}{T_t}}{2k}) \times \\
& [-\epsilon\epsilon_e \cosh(\frac{\epsilon_e}{kT_t}) + kT_t(\epsilon+\epsilon_e) \sinh(\frac{\epsilon_e}{kT_t})] - \\
& \{T \exp(\frac{\frac{bm^2}{T} + \frac{2\mu}{T}}{2k}) \times [-\epsilon\epsilon_e \cosh(\frac{\epsilon_e}{kT}) + kT(\epsilon+\epsilon_e) \sinh(\frac{\epsilon_e}{kT})] \}. \quad (18)
\end{aligned}$$

Now the numerical applications of the enthalpy and Gibbs energy increments will be taken under consideration. The unfolding and then folding of the protein molecules take place by a thermodynamical mechanism. In other words, the original structures of the macromolecules are determined thermodynamically. In the following figures, dashed lines represent experimental study where as solid lines theoretical results. The physical quantities ϵ , b and μ of the graphs which have been obtained in this semi phenomenological theory are determined by fitting to the experimental results of Privalov [1, 2]. In Figs. (1) and (2) the variations of the thermodynamical functions enthalpy and Gibbs energy increments with respect to temperature are represented for two of the selected proteins. Yellow (belonging to Myoglobin) and red (belonging to Ribonuclease) solid lines indicate the variations with respect to temperature of increments of the thermodynamical quantities which have been obtained theoretically.

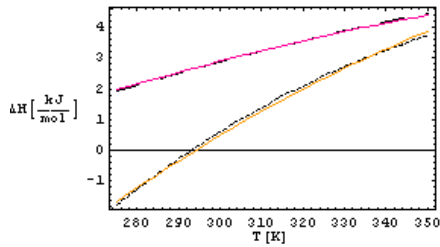


Figure 1: The variation of the additional enthalpy with temperature.

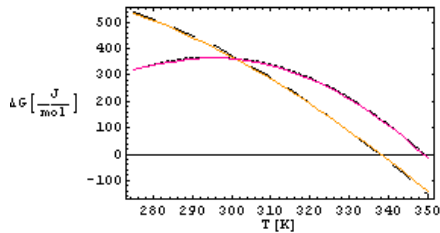


Figure 2: The variation of the additional Gibbs energy with temperature.

4 Conclusions

The enthalpy increments ΔH between the folding and unfolding states, has a negative value in the cold folding region. This behavior shows that cold folding has been realized by giving heat out and this is negative heat capacity, on the other hand in the $\Delta H > 0$ case the system is in the warm folding region and the heat capacity is positive.

The greatest contribution to the natural structure are provided by hydrophobic interactions. In an unfolding chain structure, hydrophobic amino-acid side chains are unfolded by a regular water molecule lattice and the entropy of the system decreases. But when the chain undergoes folding and hydrophobic side chains are collected at intermolecular regions by getting a way from water, the regular water molecules will be free and as increase in the total entropy of the system takes place.

In each protein molecule ΔG Gibbs potential increment, is a fraction of the quantity kT_{room} in the folding and unfolding states at each degrees of freedom and ΔG Gibbs potential increment is not a monotonic function of temperature. Gibbs free energy increment between the folding and unfolding

states has a maximum that is $\Delta G > 0$ and thus the unfolded form is stable and at both sides of this maximum this increment becomes negative. This means that two transitions take place. One of them is the unfolding of the proteins at high temperature where as the other is the folding of the proteins at low temperature. This phenomenon is known as cold denaturation in other words cold unfolding. As a result, in the cold denaturation transition hidden heat is negative and just opposite to this in the warm denaturation transition hidden heat has a positive value.

During the folding of the protein entropy increases that is to say in returning to the original structure of the protein it is not preferred from the point of view of energy. Therefore there must be some factors which oppose the increase in entropy (i.e. folding) and encourage on the other hand the folding of the molecules. Since covalent structure is conserved during unfolding, forces which oppose the unfolding of the protein could not be covalent interactions. Transformation of the non polar molecules into water becomes with a considerable decrease in entropy. The entropy of hydration of those non polar groups must be temperature dependent since hydration leads to an important increase in the heat capacity. The same thing is expected for the enthalpy of hydration. In fact, if we look at the not hydration effect during the transition of the non polar components from gas phase to water we observe that both enthalpy and entropy are negative and the magnitudes decrease with increasing temperature. Hence, one could consider that the hydration effect is responsible from the dissolution of the non polar compounds in water.

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